



Complete Genome Sequence of *Brevibacterium frigoritolerans* Ant232, Isolated from Antarctic Snow

Yong-Hoe Choe,^a Jong Ik Lee,^b  Mincheol Kim^a

^aDivision of Life Science, Korea Polar Research Institute, Incheon, Republic of Korea

^bDivision of Earth Science, Korea Polar Research Institute, Incheon, Republic of Korea

ABSTRACT We report the complete and circularized genome sequences of *Brevibacterium frigoritolerans* Ant232, generated using a combination of Illumina and PacBio platforms. The high-quality complete genome consists of a circular 5,586,945-bp chromosome and a 305,498-bp plasmid, with G+C contents of 40.66% and 36.8%, respectively.

Brevibacterium frigoritolerans was first isolated from arid soils of Morocco in 1967 and assigned to the genus *Brevibacterium*, which was established by Breed in 1953 (1). Many species of *Brevibacterium* are ubiquitously found in various environments, such as food, humans, saline soil, sediments, and marine environments (2–6). Several *Brevibacterium* strains have the potential for bioremediation of phorate in agricultural soils (7). Some *Brevibacterium* strains can tolerate abiotic stress, such as drought and salinity (8). However, only one complete genome sequence, that of *B. frigoritolerans* ZB201705, is available (8), which encouraged us to isolate and sequence the genome of *B. frigoritolerans* Ant232 to better understand the genetic properties of this species.

In this study, a bacterium named *B. frigoritolerans* Ant232 was isolated from a surface snow sample collected from inland Antarctica (74°48.399'S, 159°15.461'E) in November 2019. The snow meltwater was plated on 100% strength solid tryptone soy agar and incubated at 15°C for 3 weeks. Single colonies were selected based on morphology and/or pigmentation and were restreaked several times until pure cultures were obtained. The genomic DNA was extracted by harvesting cells on a plate cultured at 15°C for 5 days, using the MagAttract HMW DNA kit (Qiagen, Netherlands). After shearing of the genomic DNA to 20 kb, a long-read sequencing library was prepared using the SMRTbell template preparation kit v1.0 (PN 100-259-100; Pacific Biosciences [PacBio], Menlo Park, CA) and then sequenced in single-molecule real-time (SMRT) cells with PacBio RS II SMRT technology. A paired-end (2 × 150-bp) sequencing library was prepared using the Illumina TruSeq Nano DNA kit and then sequenced on the NovaSeq 6000 platform (Illumina, USA). The 664,123 total reads (read N_{50} 10,132 bp) produced by the PacBio RS II platform were assembled, whereas the Illumina platform produced 41,636,453 total reads. *De novo* assembly was performed by Flye v2.8.3 (9). The circularity of the genome and plasmid was confirmed by Circlator v1.5.5 (10). The generated assembly was quality assessed using BUSCO v4.1.2 (11), and the prediction of protein-coding sequences (CDSs) was performed using Prokka v1.14.5 (12). The functional annotation was conducted by DIAMOND v0.9.30 (13) with Gene Ontology using Blast2GO v4.1.9 (14). All tools were run with default parameters unless otherwise specified. The genome sequence of *B. frigoritolerans* Ant232 comprises a single 5,586,945-bp circular chromosome with a G+C content of 40.66% (814-fold coverage) and a single 305,498-bp circular plasmid with a G+C content of 36.8% (846-fold coverage). Annotation revealed 5,711 coding DNA sequences and 124 RNA genes (39 copies of rRNAs and 85 tRNAs). Analysis of pairwise similarities between full-length 16S rRNA gene sequences revealed that Ant232 has 99.73% sequence similarity to *B. frigoritolerans* DSM

Editor Julia A. Maresca, University of Delaware

Copyright © 2022 Choe et al. This is an open-access article distributed under the terms of the [Creative Commons Attribution 4.0 International license](https://creativecommons.org/licenses/by/4.0/).

Address correspondence to Mincheol Kim, mincheol@kopri.re.kr.

The authors declare no conflict of interest.

Received 16 February 2022

Accepted 18 March 2022

Published 7 April 2022

TABLE 1 Genome characteristics of *B. frigoritolerans* Ant232

Feature	Data for:	
	Chromosome	Plasmid
Genome size (bp)	5,586,945	305,498
Genome coverage (×)	814	846
G+C content (%)	40.66	36.8
No. of CDSs	5,389	322
No. of rRNAs	39	0
No. of tRNAs	85	0
Circular ^a	T	T
GenBank accession no.	CP084539	CP084540

^a Circular sequence: T, true; F, false.

8801. Information on the sequenced *B. frigoritolerans* Ant232 is summarized in Table 1. Average nucleotide identity (ANI) was calculated using the ANI calculator (<https://www.ezbiocloud.net/tools/ani>), which indicated that Ant232 was closely related to *B. frigoritolerans* ZB201705 (GenBank accession number [GCA_004006475.1](https://doi.org/10.1093/nar/gkz111)) (97.26%), DSM 8801 (GenBank accession number [LUUS00000000](https://doi.org/10.1093/nar/gkz111)) (97.01%), and FJAT-20673 (GenBank accession number [LWJH00000000](https://doi.org/10.1093/nar/gkz111)) (96.94%). The genome sequence of *B. frigoritolerans* Ant232 may provide a valuable resource for better understanding the mechanisms of adaptation to challenging environments.

Data availability. The complete genome sequence of *B. frigoritolerans* Ant232 has been deposited in GenBank under the accession numbers [CP084539](https://doi.org/10.1093/nar/gkz111) (genome) and [CP084540](https://doi.org/10.1093/nar/gkz111) (plasmid). The PacBio and Illumina raw data are accessible from the SRA under the accession numbers [SRR18010266](https://doi.org/10.1093/nar/gkz111) (PacBio) and [SRR18010265](https://doi.org/10.1093/nar/gkz111) (Illumina).

ACKNOWLEDGMENT

This work was supported by a grant from the Korea Polar Research Institute (grant KOPRI-PE21070).

REFERENCES

- Skerman VBD, McGowan V, Sneath PHA. 1980. Approved lists of bacterial names. *Int J Syst Evol Microbiol* 30:225–420. <https://doi.org/10.1099/00207713-30-1-225>.
- Choi EJ, Lee SH, Jung JY, Jeon CO. 2013. *Brevibacterium jeotgali* sp. nov., isolated from jeotgal, a traditional Korean fermented seafood. *Int J Syst Evol Microbiol* 63:3430–3436. <https://doi.org/10.1099/ijs.0.049197-0>.
- Wauters G, Avesani V, Laffineur K, Charlier J, Janssens M, Boesterhaut BV, Delmée M. 2003. *Brevibacterium lutescens* sp. nov., from human and environmental samples. *Int J Syst Evol Microbiol* 53:1321–1325. <https://doi.org/10.1099/ijs.0.02513-0>.
- Tang SK, Wang Y, Lou K, Mao PH, Xu LH, Jiang CL, Kim CJ, Li WJ. 2009. *Kocuria halotolerans* sp. nov., an actinobacterium isolated from a saline soil in China. *Int J Syst Evol Microbiol* 59:1316–1320. <https://doi.org/10.1099/ijs.0.006627-0>.
- Lee SD. 2006. *Brevibacterium samyangense* sp. nov., an actinomycete isolated from a beach sediment. *Int J Syst Evol Microbiol* 56:1889–1892. <https://doi.org/10.1099/ijs.0.64269-0>.
- Lee SD. 2008. *Brevibacterium marinum* sp. nov., isolated from seawater. *Int J Syst Evol Microbiol* 58:500–504. <https://doi.org/10.1099/ijs.0.65099-0>.
- Jariyal M, Gupta VK, Mandal K, Jindal V. 2015. *Brevibacterium frigoritolerans* as a novel organism for the bioremediation of phorate. *Bull Environ Contam Toxicol* 95:680–686. <https://doi.org/10.1007/s00128-015-1617-2>.
- Zhang C, Li X, Yin L, Liu C, Zou H, Wu Z, Zhang Z. 2019. Analysis of the complete genome sequence of *Brevibacterium frigoritolerans* ZB201705 isolated from drought-and salt-stressed rhizosphere soil of maize. *Ann Microbiol* 69:1489–1496. <https://doi.org/10.1007/s13213-019-01532-0>.
- Kolmogorov M, Yuan J, Lin Y, Pevzner PA. 2019. Assembly of long, error-prone reads using repeat graphs. *Nat Biotechnol* 37:540–546. <https://doi.org/10.1038/s41587-019-0072-8>.
- Hunt M, De Silva N, Otto TD, Parkhill J, Keane JA, Harris SR. 2015. Circlator: automated circularization of genome assemblies using long sequencing reads. *Genome Biol* 16:10. <https://doi.org/10.1186/s13059-015-0849-0>.
- Simão FA, Waterhouse RM, Ioannidis P, Kriventseva EV, Zdobnov EM. 2015. BUSCO: assessing genome assembly and annotation completeness with single-copy orthologs. *Bioinformatics* 31:3210–3212. <https://doi.org/10.1093/bioinformatics/btv351>.
- Seemann T. 2014. Prokka: rapid prokaryotic genome annotation. *Bioinformatics* 30:2068–2069. <https://doi.org/10.1093/bioinformatics/btu153>.
- Buchfink B, Reuter K, Drost HG. 2021. Sensitive protein alignments at tree-of-life scale using DIAMOND. *Nat Methods* 18:366–368. <https://doi.org/10.1038/s41592-021-01101-x>.
- Conesa A, Götz S, García-Gómez JM, Terol J, Talón M, Robles M. 2005. Blast2GO: a universal tool for annotation, visualization and analysis in functional genomics research. *Bioinformatics* 21:3674–3676. <https://doi.org/10.1093/bioinformatics/bti610>.